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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
10/680,286	10/06/2003	Marguerite A. Cervin	CL2180USNA	5853

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EXAMINER

CHOWDHURY, IQBAL HOSSAIN

ART UNIT	PAPER NUMBER
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1652

DATE MAILED: 07/14/2006

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/680,286	CERVIN ET AL.	
	Examiner	Art Unit	
	Iqbal Chowdhury, Ph.D.	1652	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

- 1) ☒ Responsive to communication(s) filed on 24 April 2006.
- 2a) ☐ This action is **FINAL**. 2b) ☒ This action is non-final.
- 3) ☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

- 4) ☒ Claim(s) 1-8 is/are pending in the application.
- 4a) Of the above claim(s) 4-7 is/are withdrawn from consideration.
- 5) ☐ Claim(s) _____ is/are allowed.
- 6) ☒ Claim(s) 1-3 and 8 is/are rejected.
- 7) ☐ Claim(s) _____ is/are objected to.
- 8) ☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

- 9) ☐ The specification is objected to by the Examiner.
- 10) ☐ The drawing(s) filed on _____ is/are: a) ☐ accepted or b) ☐ objected to by the Examiner.
Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).
Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).
- 11) ☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

- 12) ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).
- a) ☐ All b) ☐ Some * c) ☐ None of:
1. ☐ Certified copies of the priority documents have been received.
 2. ☐ Certified copies of the priority documents have been received in Application No. _____.
 3. ☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

- | | |
|--|---|
| 1) <input checked="" type="checkbox"/> Notice of References Cited (PTO-892) | 4) <input type="checkbox"/> Interview Summary (PTO-413) |
| 2) <input type="checkbox"/> Notice of Draftsperson's Patent Drawing Review (PTO-948) | Paper No(s)/Mail Date. _____ |
| 3) <input checked="" type="checkbox"/> Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) | 5) <input type="checkbox"/> Notice of Informal Patent Application (PTO-152) |
| Paper No(s)/Mail Date <u>02/05, 06/06</u> | 6) <input type="checkbox"/> Other: _____ |

DETAILED ACTION

This application is a non-provisional application of provisional application 60/416,192 of 10/4/2002.

The preliminary amendment filed on 4/24/2006 amending claim 8 is acknowledged. Claims 1-8 are at issue and are present for examination.

Applicant's election with traverse of Group I, Claims 1-5 and 8, drawn to an E. coli strain comprising a disrupted endogenous phosphoenolpyruvate-glucose phosphotransferase system proteins, an upregulated galP gene encoding active galactose-proton symporter, an upregulated endogenous glk gene and down regulated endogenous gapA gene and SEQ ID NO: 68 and arcA gene as species in the response filed on 4/24/2006 is acknowledged.

The amendments of claim 8 i.e. canceling SEQ ID NOS: 65-67 renders the traverse of sequence restriction moot.

The traversal is among species of genes from the following groups: ptsH gene, ptsI gene, crr gene, arcA gene, ppc gene, btuR gene, yqhD gene, mgsA gene, ackA gene, pta gene, aldA gene, aldB gene, edd gene, glpK gene, and gldA gene on the ground(s) that all the species are being capable of use together in an E. coli strain and therefore not unrelated and would not impose any burden on the examiner. Therefore, the requirement for election of species should be withdrawn. This is not found persuasive because each of the species genes is structurally and functionally independent and distinct, and searching all the species together with PEP-glucose phosphotransferase system would create a serious burden to the office. As restriction is clearly permissible even among related inventions as defined in MPEP 808 and 35 U.S.C. 121 allows

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restriction of inventions, which are independent or distinct.

The requirement is still deemed proper and is therefore made FINAL.

Claims 4, 5, 6 and 7 are withdrawn from further consideration pursuant to 37 CFR 1.142(b), as being drawn to a nonelected invention, there being no allowable generic or linking claim. Applicant timely traversed the restriction (election) requirement in communication filed on 4/24/2006.

Claims 1-3 and 8 are under consideration and are being examined herein.

Priority

Acknowledgement is made of applicants claim for priority of provisional application 60/416,192 of 10/4/2002.

Information Disclosure Statement

The information disclosure statements (IDS) submitted on 2/25/2005 and 6/19/2006 is acknowledged. The submission is in compliance with the provisions of 37 CFR 1.97. Accordingly, the information disclosure statements are being considered by the examiner.

Claim Objections

Claim 3 is objected to as encompassing non-elected subject matter. Appropriate correction is required.

Claims 3 and 8 are objected to with the recitation "further comprising" should delete "further. Appropriate correction is required.

Claim Rejections - 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter, which the applicant regards as his invention.

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Claim 8 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite and vague for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claim 8 is indefinite in the recitation “a 1.6 long GI promoter”, which is confusing. Does the term “1.6 long” refer to “1.6 kB long”? What is GI? Clarification is required.

Claim 8 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite and vague for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claim 8 is indefinite in the recitation “a 1.5 long GI promoter”, which is confusing. Does the term “1.5 long” refer to “1.5 kB long”? What is GI? Clarification is required.

The following is a quotation of the first paragraph of 35 U.S.C. 112:

The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1-3 and 8 are rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

These claims are directed to an E. coli strain comprising highly variable genus of genes including: a) disrupted endogenous phosphoenolpyruvate-glucose phosphotransferase system preventing expression of active PEP-glucose phosphotransferase system proteins; b) an up regulated endogenous galP gene encoding active galactose-proton symporter or galactose

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permease; c) an up regulated endogenous glk gene encoding active glucokinase; and d) a down regulated endogenous gapA gene encoding active glyceraldehyde 3-phosphate dehydrogenase.

Claim 2 recites the E. coli strain, wherein the disrupted endogenous phosphoenolpyruvate-glucose phosphotransferase system comprises one or more of: i) disrupted endogenous ptsH gene preventing expression of active phosphocarrier protein; ii) disrupted endogenous ptsI gene preventing expression of active phosphoenolpyruvate-protein phosphotransferase; and iii) disrupted endogenous crr gene preventing expression of active glucose-specific IIA component and Claim 3 recites E. coli strain further comprising a disrupted endogenous arcA gene preventing expression of active aerobic respiration control protein. In addition to the limitations of claims 1 and 3, claim 8 recites an E. coli strain comprising one plasmid comprising a first operon comprising genes encoding glycerol-3-phosphate dehydrogenase and glycerol-3-phosphatase, second operon further comprising 1.6 long GI promoter controlling genes encoding dehydratase and a first subunit of dehydratase reactivation factor, having sequence of SEQ ID NO: 68 that comprises orfW.

As discussed in the written description guidelines the written description requirement for a claimed genus may be satisfied through sufficient description of a representative number of species by actual reduction to practice, reduction to drawings, or by disclosure of relevant, identifying characteristics, i.e., structure or other physical and/or chemical properties, by functional characteristics coupled with a known or disclosed correlation between function and structure, or by a combination of such identifying characteristics, sufficient to show the applicant was in possession of the claimed genus. A representative number of species means that the species, which are adequately described are representative of the entire genus. Thus, when there

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is substantial variation within the genus, one must describe a sufficient variety of species to reflect the variation within the genus.

The specification teaches the structure of only single representative species of such DNAs for each genus. Moreover, the specification fails to describe any other representative species by any identifying characteristics or properties other than the functionality of encoding the recited genes. Furthermore, the specification fails to describe a method of achieving disruption of endogenous PEP-PTS system, down regulation of endogenous gapA gene, up regulation of glK and galP genes or disruption of arcA gene beyond phage transduction disruption method, putting strong promoter for up regulation or replacing ATG start codon with GTG or TTG for down regulation to produce an inactive protein system or protein or lack of protein expression due to disruption to show that applicant was in possession of the claimed genus. The specification fails to describe sufficient information to put one of skill in the art in possession of the claimed invention. Therefore, one skilled in the art cannot reasonably conclude that the applicant had possession of the claimed invention at the time the instant application was filed.

Applicant is referred to the revised guidelines concerning compliance with the written description requirement of U.S.C. 112, first paragraph, published in the Official Gazette and also available at www.uspto.gov.

Claims 1-3 and 8 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for an E. coli strain KLpts7 comprising a) disrupted endogenous phosphoenolpyruvate-glucose phosphotransferase system (operon) by using P1 phage transduction of kanamycin antibiotic resistance marker which replaces the operon genes

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comprising ptsH, ptsI and crr with the said kanamycin resistance marker; and b) an up regulated endogenous galP gene, which is under strong trc promoter, encoding active galactose-proton symporter; c) an up regulated endogenous glk gene, which is under strong trc promoter, encoding active glucokinase; and d) a down regulated endogenous gapA gene encoding active glyceraldehyde 3-phosphate dehydrogenase by replacing ATG start codon with GTG or TTG and disrupted endogenous arcA gene by using pKD3 gene knockout system for preventing expression of active aerobic respiration control protein and further comprising one plasmid comprising a first operon comprising genes encoding glycerol-3-phosphate dehydrogenase and glycerol-3-phosphatase, second operon further comprising 1.6 long GI promoter controlling genes encoding dehydratase and a first subunit of dehydratase reactivation factor, having sequence of SEQ ID NO: 68, does not reasonably provide enablement for any E. coli strain comprising a) any disrupted endogenous phosphoenolpyruvate-glucose phosphotransferase system (operon); and b) up regulation of any endogenous galP gene; c) up regulation of any endogenous glk gene; and d) a down regulation of any endogenous gapA gene and disruption of any endogenous arcA gene and further comprising any glycerol-3-phosphate dehydrogenase gene and any glycerol-3-phosphatase gene. The specification does not enable any person skilled in the art to which it pertains; or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims.

Claims 1-3 and 8 are so broad as to encompass any E. coli strain comprising: a) a disrupted endogenous phosphoenolpyruvate-glucose phosphotransferase system (operon); and b) an up regulated any endogenous galP gene; c) an up regulated endogenous glk gene; and d) a down regulated endogenous gapA gene and a disruption of the endogenous arcA gene and

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further comprising any glycerol-3-phosphate dehydrogenase gene and any glycerol-3-phosphatase gene for the production of 1,3-propanediol from glucose. The scope of the claims is not commensurate with the enablement provided by the disclosure with regard to the extremely large number of E. coli strains encompassed as the claims do not specify how the endogenous E. coli Pts, galP, gapA, arcA, G3PDH and glycerol 3-phosphatase genes are modified to produce the up/down regulated genes recited. Since the amino acid sequence of a protein determines its structural and functional properties, predictability of which changes can be tolerated in a protein's amino acid sequence and obtain the desired activity requires a knowledge of and guidance with regard to which amino acids in the protein's sequence, if any, are tolerant of modification and which are conserved (i.e. expectedly intolerant to modification), and detailed knowledge of the ways in which the proteins' structure relates to its function. However, in this case the disclosure is limited to the nucleotide and encoded amino acid sequences of only a few recombinant E. coli strains useful within the claimed methods.

The scope of the claims is not commensurate with the enablement provided by the disclosure with regard to the methods of modification (disruption) of E. coli endogenous PEP-PTS genes (ptsH gene, ptsI gene, and crr gene) or arcA gene for modified expression or reduced expression of gapA gene or increased expression of galP and glk gene broadly encompassed by the claims. Since the expression of a protein determines its structural and functional properties, predictability of which changes can be tolerated in a protein's amino acid sequence or in the promoter sequence and obtain the desired activity requires a knowledge of and guidance with regard to which amino acids in the protein's sequence or the promoter sequence and techniques if any, are tolerant of modification, and detailed knowledge of the ways in which the proteins'

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expression relates to its function. However, in this case the disclosure is limited to the modification of PEP-PTS system and *arcA* gene by P1 phage transduction (disruption), up regulation of *glk* and *galP* genes by putting strong *Trc* promoter and down regulating endogenous *gapA* gene by replacing ATG start codon with GTG or TTG codon.

The specification also does not support the broad scope of the claims, which encompass methods of using an *E. coli* comprising any disrupted PEP-PTS genes or *arcA* gene, down regulating of any *gapA* gene or up regulating of any *galP* gene or *glk* gene because the specification does not establish: (A) regions of the protein structure which may be modified such that PEP-PTS system proteins and *arcA* gene encoding protein lacks activity and reduced activity of *gapA* gene encoding protein and up regulating activity of *galP* or *glk* gene encoding proteins; (B) the general tolerance of *gapA* or *galP* or *glk* proteins to modification and extent of such tolerance; (C) a rational and predictable scheme for modifying any PEP-PTS system proteins, *arcA* protein, *gapA* protein, *galP* or *glk* protein residues with an expectation of obtaining the desired biological function; and (D) the specification provides insufficient guidance as to which of the essentially infinite possible choices is likely to be successful.

Thus, applicants have not provided sufficient guidance to enable one of ordinary skill in the art to make and use the claimed invention in a manner reasonably correlated with the scope of the claims broadly including any *E. coli* strain comprising: a) a disrupted endogenous phosphoenolpyruvate-glucose phosphotransferase system (operon); and b) an up regulated endogenous *galP* gene; c) an up regulated endogenous *glk* gene; and d) a down regulated endogenous *gapA* gene and disruption of the endogenous *arcA* gene and further comprising any glycerol-3-phosphate dehydrogenase gene and any glycerol-3-phosphatase genes.

In addition, applicants have not provided sufficient guidance to enable one of ordinary skill in the art to make and use the claimed invention in a manner reasonably correlated with the scope of the claims broadly including methods of disrupting any PEP-ETS system genes or arcA genes or methods of down regulating expression of gapA gene or up regulating of any galP or glk genes.

The scope of the claims must bear a reasonable correlation with the scope of enablement (In re Fisher, 166 USPQ 19 24 (CCPA 1970)). Without sufficient guidance, determination of any E. coli strain comprising: a) disrupted any endogenous phosphoenolpyruvate-glucose phosphotransferase system (operon); and b) up regulated any endogenous galP gene; c) up regulated any endogenous glk gene; and d) a down regulated any endogenous gapA gene and disruption of any endogenous arcA gene and further comprising any glycerol-3-phosphate dehydrogenase gene and any glycerol-3-phosphatase genes to use in the claimed methods having the desired biological characteristics is unpredictable and the experimentation left to those skilled in the art is unnecessarily, and improperly, extensive and undue. See In re Wands 858 F.2d 731, 8 USPQ2nd 1400 (Fed. Cir, 1988).

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

(a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negated by the manner in which the invention was made.

The factual inquiries set forth in *Graham v. John Deere Co.*, 383 U.S. 1, 148 USPQ 459 (1966), that are applied for establishing a background for determining obviousness under 35 U.S.C. 103(a) are summarized as follows:

1. Determining the scope and contents of the prior art.
2. Ascertaining the differences between the prior art and the claims at issue.
3. Resolving the level of ordinary skill in the pertinent art.
4. Considering objective evidence present in the application indicating obviousness or nonobviousness.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

Claims 1-3 and 8 are rejected under 35 U.S.C. 103(a) as being unpatentable over Baez et al. (Determination of 3-deoxy-D-arabino-heptulosonate 7-phosphate productivity and yield from glucose in *Escherichia coli* devoid of the glucose phosphotransferase transport system. *Biotechnol Bioeng.* 2001 Jun 20; 73(6): 530-5), Seta et al. (Characterization of *Escherichia coli* strains with gapA and gapB genes deleted, *J. Bacteriol.* 1997 Aug; 179(16): 5218-21), Iuchi et al. (arcA (dye), a global regulatory gene in *Escherichia coli* mediating repression of enzymes in aerobic pathways, *Proc Natl Acad Sci U S A.* 1988 Mar; 85(6): 1888-92) in view of Emptage et al. (WO 01/12833 A2, publication 2/22/2001, Process for the biological production of 1,3-

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propanediol with high titer, see IDS) and Payne et al. (US PG PUB 20050147968 A1, publication 7/7/2005, claim priority of 60/374931 of 4/22/2002).

Baez et al. teach a recombinant *E. coli* strain devoid of the glucose phosphotransferase transport system NF9 (PTS- glucose+), which can utilize glucose for the production of DAHP. Baez et al. also teach the strain comprising glucokinase gene (*glk* gene), wherein the activity is higher than the parental strain, which suggest that replacement of PTS function by *galP* and *glK* gene that indicates the up regulation of *galP* gene compared to parental strain. Baez et al. do not teach *E. coli* strain comprising reduced expression of glyceraldehyde dehydrogenase (*gapA*) gene and disrupted *arcA* (global regulatory protein) gene or expression of G3PDH or G3P phosphatase genes or SEQ ID NO: 68 (pSYCO109 plasmid).

Seta et al. teach an *E. coli* strain having a deleted glyceraldehyde 3-phosphate dehydrogenase (*gapA*) gene and suggested that the essential role of *gapA* gene in glycolysis. Seta et al. do not teach *arcA* gene or expression of G3PDH or G3P phosphatase genes or SEQ ID NO: 68 (pSYCO109 plasmid).

Iuchi et al. teach a recombinant *Escherichia coli*, wherein an *arcA* gene is mutated, deleted or disrupted resulting in the increased expression of the enzymes, which are specifically express at anaerobic condition. Iuchi et al. do not teach expression of G3PDH or G3P phosphatase genes or dehydratase or dehydratase reactivation factor or *orfW* gene expression or SEQ ID NO: 68 (pSYCO109 plasmid).

Emtage et al. teach a process for the biological production of 1,3-propanediol with high titer by using a recombinant *E. coli* comprising and expressing 1,3-propanediol oxidoreductase (*dhaT*), Glycerol 3-phosphate dehydrogenase (G3PDH) or Glycerol 3-phosphate phosphatase

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(G3Pase) genes or dehydratase gene or dehydratase reactivation factor gene or orfW gene for efficient production of 1,3-propanediol. Emptage et al. do not teach E. coli comprising disrupted glucose phosphotransferase transport system NF9 (PTS- glucose+) or arcA gene or up regulating galP and glk genes or reduced expression of gapA gene or SEQ ID NO: 68 (pSYCO109 plasmid).

Payne et al. disclose SEQ ID NO: 68 or pSYCO109 plasmid comprising the enzymes expressed by said plasmid including glycerol dehydratase, dehydratase reactivation factor, glycerol-3-phosphate dehydrogenase, and glycerol-3-phosphatase in E. coli RJ8n strain. Payne et al. also teach GI promoter including 1.5 and 1.6; yqhD gene, and orfW gene.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to combine the teachings of Baez et al., Seta et al., Iuchi et al., Emptage et al. and Payne et al. to construct a recombinant E. coli strain disrupted PTS system genes as taught by Baez et al. deleted gapA gene as taught by Seta et al., deleted arcA gene as disclosed by Iuchi et al. and over-expression of galP gene, glk gene, G3PDH gene, G3Pase gene or dehydratase gene or dehydratase reactivation factor gene or orfW genes as taught by Emptage et al. and using SEQ ID NO: 68 (pSYCO109 plasmid), which includes glycerol dehydratase, dehydratase reactivation factor, glycerol-3-phosphate dehydrogenase, and glycerol-3-phosphatase in E. coli RJ8n strain. Payne et al. also teach GI promoter including 1.5 and 1.6; yqhD gene, and orfW gene as taught by Payne et al. for the production of 1,3-propanediol, and do so with a reasonable expectation of success.

Conclusion

Status of the claims:

Claims 1-3 and 8 are pending.

Claims 1-3 and 8 are rejected.

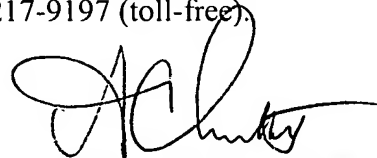
No claim is in condition for allowance.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Iqbal Chowdhury whose telephone number is 571-272-8137. The examiner can normally be reached on 9:00-5:00 PM.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ponnathapu Achutamurthy can be reached on 703-272-0928. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see <http://pair-direct.uspto.gov>. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

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